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A new interpretation of the σ_A parameter

B. Carrozzini, G. L. Cascarano, C. Giacovazzo* and A. Mazzone

Istituto di Cristallografia, CNR, Via Amendola 122/o, Bari, Italy. Correspondence e-mail: carmelo.giacovazzo@ic.cnr.it

A new study of the σ_A parameter has been undertaken to understand its behaviour when the diffraction amplitude distributions are far from the standard Wilson distributions. The study has led to the formulation of a new statistical interpretation of σ_A, expressed in terms of a correlation factor. The new formulas allow a more accurate use of σ_A in electron-density modification procedures.

1. Notation

N, N_p, number of atoms in the unit cell of the target and of the model structure, respectively. Usually $N_p \leq N$, but it may also be $N_p > N$.
f_j, $j = 1, \ldots, N$, atomic scattering factors for the target structure (thermal factor included).
$F = \sum_{j=1}^{N} f_j \exp(2\pi i \mathbf{h}\mathbf{r}_j) = |F| \exp(i\varphi)$, structure factor of the target structure.
$F_p = \sum_{j=1}^{N_p} f_j \exp(2\pi i \mathbf{h}\mathbf{r}'_j) = |F_p| \exp(i\varphi_p)$, structure factor of the model structure. The atomic positions of the model structure are related to those of the target by the relation $\mathbf{r}'_j = \mathbf{r}_j + \Delta\mathbf{r}_j$.
$\Sigma_N = \sum_{j=1}^{N} f_j^2$, $\Sigma_{N_p} = \sum_{j=1}^{N_p} f_j^2$.
$E = F/(\Sigma_N)^{1/2} = R\exp(i\varphi)$, $E_p = F/(\Sigma_{N_p})^{1/2} = R_p\exp(i\varphi_p)$, normalized structure factors of F and F_p, respectively. R and R_p are the moduli of E and E_p, respectively.
$D = \langle \cos 2\pi \mathbf{h}\Delta\mathbf{r} \rangle$, $\langle \Delta\mathbf{r} \rangle$ is the average vectorial difference between the N_p positional vectors of the model atoms and the corresponding vectors in the target structure.

$$\sigma_A = D(\Sigma_{N_p}/\Sigma_N)^{1/2} \quad \text{or} \quad \sigma_A = D(\Sigma_N/\Sigma_{N_p})^{1/2} \qquad (1)$$

according to whether Σ_{N_p} is smaller or larger than Σ_N.
$\sigma_R^2 = \langle |\mu|^2 \rangle / \Sigma_N$, $\langle |\mu|^2 \rangle$ is the measurement error and σ_R^2 is its normalized form.
$e = 1 + \sigma_R^2$.
$I_i(X)$, modified Bessel function of order i.
$D_i(X) = I_i(X)/I_0(X)$.
EDM, electron-density modification.
Paper I, Burla, Giacovazzo *et al.* (2011).

2. Introduction

Hauptman (1982), in his theory integrating direct-methods techniques with isomorphous replacement and in accordance with Sim's (1959) assumptions, supposed that the atomic positions $\mathbf{r}_j$ for the two isomorphous structures were the same: f_j are the scattering factors in the target structure, g_j in the model structure. When $g_j = 0$, no atom is in the jth position. Hauptman's theory establishes that

$$\frac{\langle R^2 R_p^2 \rangle - \langle R^2 \rangle \langle R_p^2 \rangle}{(\langle R^4 \rangle - \langle R^2 \rangle^2)^{1/2} (\langle R_p^4 \rangle - \langle R_p^2 \rangle^2)^{1/2}} = \alpha^2,$$

where

$$\alpha^2 = \frac{(\sum_j f_j g_j)^2}{(\sum_j f_j^2)(\sum_j g_j^2)}.$$

If $f_j = g_j$, unless $g_j = 0$ when no atom is in the position $\mathbf{r}_j$, then

$$\alpha^2 = \Sigma_p / \Sigma_N.$$

Luzzati (1952) started the study of isomorphous structures, where corresponding atoms show a misfit in the atomic positions. The corresponding $P(E, E_p)$ distribution was provided by Srinivasan & Ramachandran (1965), who greatly enlarged the concept of model and target structure and started the study of the parameter

$$\sigma_A^2 = D^2 \Sigma_p / \Sigma_N.$$

In accordance with Srinivasan and Ramachandran, in our approach the atomic positions in the target and in the model may differ by $\Delta\mathbf{r}_j$.

The role of σ_A in modern phasing techniques rapidly increased, as the following examples suggest:

(*a*) Read (1986) used the likelihood function given by Lunin & Urzhumtsev (1984) to provide the probability of the observed structure factors when a model structure is available. The corresponding weight is

$$m = D_1(X) \quad \text{or} \quad m = \tanh(X/2) \qquad (2)$$

with

$$X = 2\sigma_A R R_p / (1 - \sigma_A^2) \qquad (3)$$

according to whether the crystal is acentric or centric.

(*b*) Equations (2) were generalized by Caliandro *et al.* (2005) to take into account the measurement error; they are still valid but X should be modified into

$$X = 2\sigma_A R R_p / (e - \sigma_A^2). \qquad (4)$$

(*c*) The equations (2)–(4) are widely used in EDM procedures.

(*d*) The accurate σ_A estimate is crucial for the *VLD* method (Burla, Carrozzini *et al.*, 2011; Burla, Carrozzini, Cascarano, Giacovazzo & Polidori, 2012), a new *ab initio* phasing approach using the properties of the Fourier transform for recovering the correct structure from a random model.

(*e*) More recently, the parameter has been involved in a formula estimating triplet phase invariants given a model structure (Burla, Carrozzini, Cascarano, Comunale *et al.*, 2012).

(*f*) In computer programs refining macromolecular structures by maximum-likelihood methods (*e.g.* see *REFMAC* by Murshudov *et al.*, 1997), σ_A may be estimated from reflections not included in the refinement of atomic parameters and is also used for restoring unmeasured reflections in electron-density syntheses.

In order to estimate σ_A, the measured reflections are partitioned in resolution shells (indeed σ_A is a resolution-dependent parameter) and for each shell the mixed moments $\langle R^2 R_p^2\rangle$ are calculated, from which

$$\sigma_A^2 = (\langle R^2 R_p^2\rangle - e) \tag{5a}$$

or

$$\sigma_A^2 = \tfrac{1}{2}(\langle R^2 R_p^2\rangle - e) \tag{5b}$$

for acentric or centric crystals, respectively.

To reduce the effects of the experimental structure-factor distributions, when not coincident with Wilson distributions, in practical applications [*e.g.* in the program *SIGMAA* by Read (1986)] it is usual to renormalize the structure factors per shell. This is equivalent to using

$$\sigma_A^2 = \left(\frac{\langle R^2 R_p^2\rangle}{\langle R^2\rangle\langle R_p^2\rangle} - e\right) \quad \text{and} \quad \sigma_A^2 = \frac{1}{2}\left(\frac{\langle R^2 R_p^2\rangle}{\langle R^2\rangle\langle R_p^2\rangle} - e\right) \tag{6}$$

for acentric and centric crystals, respectively. In terms of structure factors the above equations may be rewritten as

$$\sigma_A^2 = \left(\frac{\langle |FF_p|^2\rangle}{\langle |F|^2\rangle\langle |F_p|^2\rangle} - e\right) \text{ and } \sigma_A^2 = \frac{1}{2}\left(\frac{\langle |FF_p|^2\rangle}{\langle |F|^2\rangle\langle |F_p|^2\rangle} - e\right),$$

respectively.

Applying equations (6) instead of equations (5*a*) and (5*b*) does not have a theoretical basis, but it helps in practice to reduce the effects of the non-Wilsonian amplitude distributions. It is, however, obvious that the renormalization *via* the moments of order two used in equations (6) cannot obviate the distortions of the σ_A estimate generated by the non-Wilsonian nature of the actual distributions (indeed, by definition, each distribution is defined by the full set of its moments, not only by moments of order two).

The above-described state of the art is therefore not satisfactory. Indeed:

(i) In direct space the definition of equation (1) exactly defines the σ_A parameter in terms of the incompleteness of the model (say the ratio Σ_{N_p}/Σ_N) and of the similarity between the model and the target structure (say *via* the *D* term). In reciprocal space (the space in which σ_A is estimated) a statistical interpretation of σ_A is still not available.

(ii) The mathematical relationships established through equations (2)–(5*a*),(5*b*) are no longer valid if the local and/or the global amplitude distribution do not fit the Wilson statistics. As stated before, the renormalization *via* moments of order two cannot capture the essential features of the non-Wilsonian distributions; as a consequence, overestimation or underestimation of the σ_A values may occur, with possible loss of efficiency of the EDM procedures. It often occurs that the reciprocal-space averages by means of which σ_A is estimated provide values larger than unity.

A new study is therefore useful to estimate σ_A when the diffraction amplitude distributions are far from the Wilson standards (for a different approach see paper I). This is one of the two purposes of this paper: the second, correlated with the first, is to identify a new statistical interpretation of σ_A, characterized by a more general mathematical expression.

3. A new σ_A interpretation

In order to make the following calculations immediately readable, let us assume that $e = 1$ (this approximation is practically satisfied for all the largest R values).

From the classical Srinivasan and Ramachandran acentric distribution the following marginal distribution is obtained by standard techniques,

$$P(R, R_p) = \frac{4}{(1-\sigma_A^2)} R R_p \exp\left\{-\frac{1}{(1-\sigma_A^2)}[R^2 + R_p^2]\right\} I_0[X], \tag{7}$$

which may be used to calculate the joint moment

$$\langle R^2 R_p^2\rangle = \int_0^\infty R^2 R_p^2 \, \mathrm{d}R \, \mathrm{d}R_p. \tag{8}$$

It is easily shown that, if the integration operations are first made over R_p and then over R,

$$\begin{aligned}\langle R^2 R_p^2\rangle &= (1-\sigma_A^2)\int_0^\infty R^3 \exp(-R^2)\,\mathrm{d}R + \sigma_A^2 \int_0^\infty R^5 \exp(-R^2)\,\mathrm{d}R \\ &= (1-\sigma_A^2)\langle R^2\rangle + \sigma_A^2\langle R^4\rangle \end{aligned} \tag{9}$$

is obtained, from which

$$\sigma_A^2 = \frac{\langle R^2 R_p^2\rangle - \langle R^2\rangle}{(\langle R^4\rangle - \langle R^2\rangle)}. \tag{10}$$

If we replace in equation (10) the fourth- and the second-order marginal moments by the Wilson expected values (say $\langle R^2\rangle = 1$ and $\langle R^4\rangle = 2$), then the relation (5*a*) is obtained.

Let us now integrate equation (8) first over R and then over R_p; we obtain

$$\langle R^2 R_p^2\rangle = (1-\sigma_A^2)\int_0^\infty R_p^3 \exp(-R_p^2)\,\mathrm{d}R + \sigma_A^2 \int_0^\infty R_p^5 \exp(-R_p^5)\,\mathrm{d}R_p$$

from which

$$\sigma_A^2 = \frac{\langle R^2 R_p^2\rangle - \langle R_p^2\rangle}{(\langle R_p^4\rangle - \langle R_p^2\rangle)}. \quad (11)$$

Again, equation (11) reduces to equation (5*a*) if the marginal moments of order two and four are replaced by their Wilson expected values.

Relations identical to equations (10) and (11) are obtained when the centric distribution $P(R, R_p)$ is taken into account. From equations (10) and (11) the relation (5*b*) is obtained by replacing the moments of order two and four by the Wilson expected values $\langle R^2\rangle = 1$ and $\langle R^4\rangle = 3$.

The above results suggest the following conclusions:

(i) If R and R_p obey the Wilson distribution, the order of integration in equation (8) is not relevant and the relationships in equations (5*a*) and (5*b*) are obtained.

(ii) If R and R_p do not obey the Wilson distribution, two different relationships are obtained from equation (8) [say equations (10) and (11)], according to the order of integration. The asymmetry in the results is the effect of a hypothesis which has been introduced in the calculations but which was not in the original bivariate distribution.

(iii) The moments of order four are necessarily involved when the integration is performed. They do not explicitly appear because they are replaced by the Wilsonian values: they should be explicitly considered in the case of non-Wilsonian distributions.

Unfortunately, the observed and calculated amplitude distributions are often non-Wilsonian; in this case the algebraic form of equations (5*a*) and (5*b*) does not hold anymore because it will depend on the specific experimental distribution.

It may then be useful to replace equations (5*a*) and (5*b*) by a more general relation which: (i) makes use of the marginal moments $\langle R^4\rangle$ and $\langle R_p^4\rangle$, as suggested by equations (10) and (11); (ii) reduces to equations (5*a*) and (5*b*) when the observed and calculated structure-factor distributions are Wilson-like; (iii) always provides estimates in the interval (0, 1). Our proposal is the following: σ_A^2 is nothing but the correlation factor C between the R^2 and the R_p^2 sets:

$$\sigma_A^2 = C(R^2, R_p^2) = \frac{\langle R^2 R_p^2\rangle - \langle R^2\rangle\langle R_p^2\rangle}{(\langle R^4\rangle - \langle R^2\rangle^2)^{1/2}(\langle R_p^4\rangle - \langle R_p^2\rangle^2)^{1/2}}. \quad (12)$$

We observe:

(*a*) σ_A^2, as defined in direct space, always lies in the interval (0, 1). The reciprocal-space estimates [equations (5*a*) and (5*b*)] may often be outside the interval (0, 1), against the definition of equation (1). Equation (12) always guarantees the correct interval.

(*b*) Equation (12) does not change with the centric or acentric nature of the crystal, and reduces to equation (5*a*) or (5*b*) if the structure factor satisfies the corresponding Wilson distribution; it is sufficient to replace the marginal moments by their expected Wilson values. That is very useful when the moments are calculated from sets containing general and symmetry-restricted reflections.

(*c*) Rescaling of the observed and/or the calculated amplitudes shell per shell, as described by equation (6), is no longer necessary because the correlation coefficient is scale independent.

(*d*) When $D = 1$ (that is when $\Delta\mathbf{r}_j = 0$ for $j = 1, \ldots, N_p$) our result coincides with the Hauptman (1982) formula reported in the first lines of §2.

The new statistical interpretation of σ_A^2 has been obtained by considering the mixed moment $\langle R^2 R_p^2\rangle$, but it may be shown that it has a more general validity. Let us consider, for example, the moment $\langle RR_p\rangle$: if both R and R_p satisfy the acentric Wilson distribution, then (Caliandro *et al.*, 2005)

$$\langle RR_p\rangle = \frac{\pi}{4}\,{}_1F_1\left(\frac{-1}{2}, \frac{-1}{2}; 1; \sigma_A^2\right), \quad (13)$$

where ${}_1F_1$ is the confluent hypergeometric function. According to paper I, equation (13) may be approximated by

$$\langle RR_p\rangle = \frac{\pi}{4} e^{1/2}\left(1 + \frac{\pi}{12}\frac{\sigma_A^2}{e}\right),$$

from which, after simple calculations,

$$\sigma_A^2 = \left(\langle RR_p\rangle - \frac{\pi}{4}\right)\Big/\left(1 - \frac{\pi}{4}\right) \quad (14)$$

is obtained. Since $\langle R\rangle = \langle R_p\rangle = (\pi)^{1/2}/2$, equation (14) may be rewritten in terms of a correlation coefficient:

$$\sigma_A^2 = C(R, R_p) = \frac{\langle RR_p\rangle - \langle R\rangle\langle R_p\rangle}{(\langle R^2\rangle - \langle R\rangle^2)^{1/2}(\langle R_p^2\rangle - \langle R_p\rangle^2)^{1/2}}. \quad (15)$$

It may be argued that σ_A^2 may be estimated *via* any moment $\langle R^n R_p^n\rangle$ in the form of correlation coefficients, and that from each moment a specific estimate arises, correlated but not identical with the others.

Does the statistical interpretation of σ_A^2 as a correlation factor have a counterpart in direct space? In other words, can σ_A^2 be interpreted in terms of correlation between the electron density of the target and of the model structure? In particular, does $C(R^2, R_p^2)$ coincide with the correlation $C(\rho, \rho_p)$ given by

$$C(\rho, \rho_p) = \frac{\langle \rho\rho_p\rangle - \langle \rho\rangle\langle \rho_p\rangle}{(\langle \rho^2\rangle - \langle \rho\rangle^2)^{1/2}(\langle \rho_p^2\rangle - \langle \rho_p\rangle^2)^{1/2}}? \quad (16)$$

Since the usual electron densities do not contain the constant coefficient, the definition of equation (16) reduces to

$$C(\rho, \rho_p) = \frac{\langle \rho\rho_p\rangle}{(\langle \rho^2\rangle)^{1/2}(\langle \rho_p^2\rangle)^{1/2}}. \quad (17)$$

In order to estimate such correlation, we need to calculate the integral

$$\int_V \rho(\mathbf{r})\rho_p(\mathbf{r})\,\mathrm{d}\mathbf{r} = \frac{1}{V}\sum_{\mathbf{h}} |F_{\mathbf{h}} F_{p\mathbf{h}}| \cos(\varphi_{\mathbf{h}} - \varphi_{p\mathbf{h}}). \quad (18)$$

In the absence of information on the target phases we can approximate equation (18) by

$$\int_V \rho(\mathbf{r})\rho_p(\mathbf{r})\,\mathrm{d}\mathbf{r} \simeq \frac{1}{V}\sum_{\mathbf{h}} |F_{\mathbf{h}}F_{p\mathbf{h}}|D_1(X_{\mathbf{h}}). \quad (19)$$

Using the well known Parseval relation

$$\int_V \rho^2(\mathbf{r})\,\mathrm{d}\mathbf{r} = \frac{1}{V}\sum_{\mathbf{h}} |F_{\mathbf{h}}|^2$$

leads to

$$C(\rho, \rho_p) = \frac{\sum_{\mathbf{h}} |F_{\mathbf{h}}F_{p\mathbf{h}}|D_1(X_{\mathbf{h}})}{\left(\sum_{\mathbf{h}} |F_{\mathbf{h}}|^2\right)^{1/2}\left(\sum_{\mathbf{h}} |F_{p\mathbf{h}}|^2\right)^{1/2}}$$

or, in terms of normalized structure factors, to

$$C(\rho, \rho_p) = \langle R_{\mathbf{h}}R_{p\mathbf{h}}D_1(X_{\mathbf{h}})\rangle. \quad (20)$$

The above results show that σ_A^2 cannot be interpreted as the correlation between the electron-density maps of the target and of the model structure. However, it influences such a correlation: large values of $\langle\sigma_A^2\rangle$ imply large $C(\rho, \rho_p)$ values.

4. Experimental tests

The observed and the calculated amplitude distributions do not always comply with the standard Wilson distributions. Among the several structural reasons which lead to distorted distributions we quote: (i) some heavy atoms are in special positions; (ii) groups of atoms have a centric (in acentric structures) or a hypercentric arrangement; (iii) in acentric structures groups of atoms are shifted by small displacements from positions compatible with the presence of an inversion centre; (iv) pseudo-translational symmetry is present.

While the last pseudo-symmetry type may be assessed by checking the statistics of the observed amplitudes (Cascarano *et al.*, 1985, 1987), the others cannot be easily diagnosed if a molecular model is not available.

The general effect of the pseudo-symmetry on the amplitude statistics is the increase of the high-order marginal moments (*e.g.* $\langle R^4\rangle$, $\langle R^6\rangle$, ...) and, often, of the mixed moments $\langle R^n R_p^n\rangle$: they become larger than expected from the Wilson distributions. The rationale is the following: the pseudo-symmetries divide the reflections into groups, each group with its own value of $\langle|F|^2\rangle$. In the absence of any information on the pseudo-symmetry, the observed and the calculated amplitudes are normalized by dividing them by the overall expected scattering power at the given resolution shell [say $(\Sigma_N)^{1/2}$ and $(\Sigma_{N_p})^{1/2}$, respectively]; as a result, some groups of reflections will show large average values of $\langle R^2\rangle$ and other groups will show smaller values of $\langle R^2\rangle$. This behaviour statistically leads to increased values of the higher-order moments $\langle R^n\rangle$ with $n > 2$ and, often, of the mixed moments $\langle R^n R_p^n\rangle$.

In general, pseudo-symmetry effects lead to overestimated σ_A values. Luckily, the interpretation of σ_A^2 as a correlation factor allows one to reduce such an effect by using the moments of order four at the denominator of equations (12) and (15), rather than the Wilson expected values.

Table 1
Test structures.

For each structure the PDB code, the space group (SG), the experimental data resolution (RES) and the moments $\langle R\rangle$ and $\langle R^4\rangle$ are given. If pseudo-symmetry is present, the pseudo-translational vector is characterized *via* the pseudo-translation vector **u** and the percentage of electron density satisfying the pseudo-translation (%).

PDB	SG	RES	$\langle R\rangle$	$\langle R^4\rangle$	**u**	%	Reference
1ick	$P2_12_12_1$	0.95	0.87	2.39			(*a*)
1nkd	$C2$	1.1	0.85	2.68			(*b*)
1j6s	$P42_12$	1.40	0.84	2.92	**c**/2	25	(*c*)
1dy5	$P2_1$	0.87	0.87	2.33	**a**/2	55	(*d*)
1lys	$P2_1$	1.72	0.90	2.15	(**a**+**c**)/2	54	(*e*)
2hyw	$P2_1$	2.1	0.79	3.54	(**b**+**c**)/2	89	(*f*)
1yxa	$P2_12_12_1$	2.1	0.86	2.44	**a**/2	20	(*g*)
2p0g	$P3_12_1$	2.3	0.83	2.78	**c**/2	75	(*h*)

References: (*a*) Dauter & Adamiak (2001); (*b*) Vlassi *et al.* (1998); (*c*) Pan *et al.* (2003); (*d*) Esposito *et al.* (2000); (*e*) Harata (1994); (*f*) Shao *et al.* (2006); (*g*) Horvath *et al.* (2013); (*h*) Benach *et al.* (2013).

In order to confirm the above statements we have considered eight test structures quoted in Table 1, six of which are affected by pseudo-translational symmetry; for all of them the experimental moment $\langle R^4\rangle$ is significantly larger than two. In the table the nature of the pseudo-translational symmetry, when present [say, the pseudo-translational vector **u** and the percentage of electron density (%) satisfying it], is reported. While $\langle R^4\rangle$ moments are close to centric of hypercentric values, the $\langle R\rangle$ moments do not show strong deviations from the Wilson acentric values. The only exception is the protein with PDB (Protein Data Bank) code 2hyw, for which $\langle R\rangle$ is close to the centric values.

To compare the σ_A estimates provided by equations (6), (12) and (15), the phase error may be monitored against the calculated σ_A values during the full phasing process. Proteins with PDB codes 1ick and 1nkd are particularly indicated for such a purpose because *ab initio* phasing techniques may be successfully applied to them [*i.e.*, a Patterson deconvolution approach followed by EDM procedures, as implemented in *SIR2011*; Burla, Caliandro *et al.* (2012)]. In this case the behaviour of the σ_A estimates provided by equations (6), (12) and (15) may be observed for a wide interval of the phase error. We calculated the σ_A values during the EDM cycles: the error varies from about 80° (typical when the heavy atoms are not so heavy, like Cl or S) up to about 25° (typical for such small high-resolution proteins).

The σ_A trend for proteins with PDB codes 1ick and 1nkd is shown in Figs. 1 and 2, respectively. We notice:

(i) The σ_A parameter is in general overestimated. This is an intrinsic feature of EDM procedures, because the calculated structure factors are obtained by modification and inversion of observed electron-density maps.

(ii) The largest overestimation is obtained by using equation (6), followed by equation (15) and by equation (12) in that order, in agreement with the theoretical expectations.

The above trend is confirmed for all the test structures. As a consequence, it may be guessed that for structures with a high $\langle R^4\rangle$ moment, the σ_A parameter is expected to attain values close to unity even when the phase error is significantly far

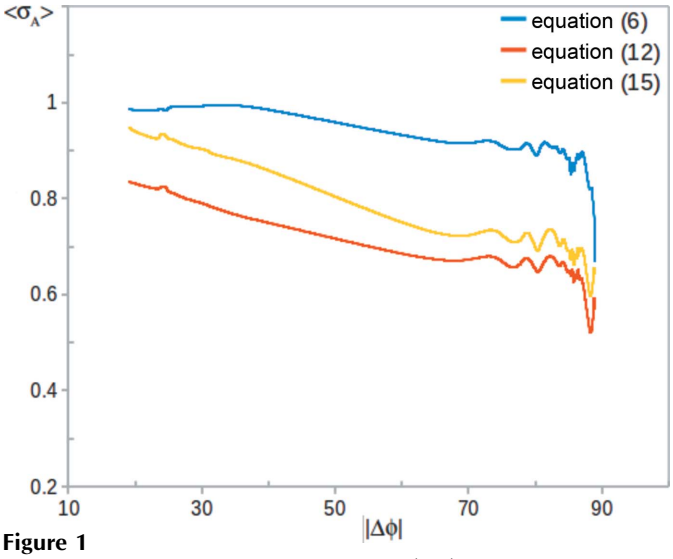

Figure 1
1ick. The σ_A estimates *versus* the phase error $|\Delta\varphi|$ as obtained by *SIR2011* when *ab initio* Patterson deconvolution techniques are applied.

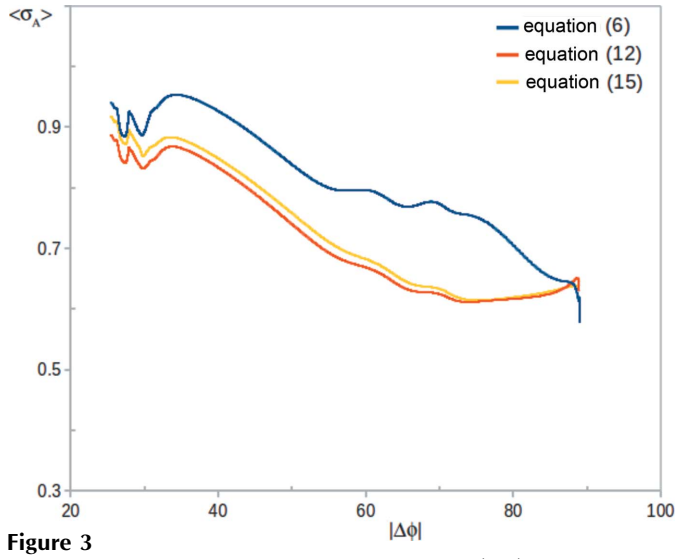

Figure 3
1a0m. The σ_A estimates *versus* the phase error $|\Delta\varphi|$ as obtained by *SIR2011* when *ab initio* Patterson deconvolution techniques are applied.

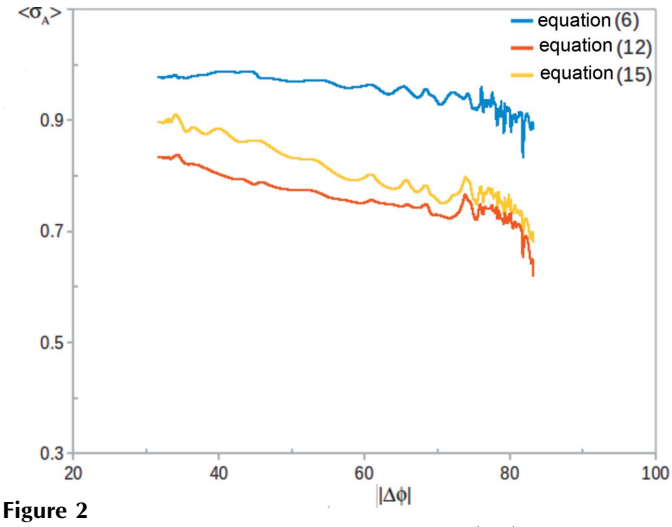

Figure 2
1nkd. The σ_A estimates *versus* the phase error $|\Delta\varphi|$ as obtained by *SIR2011* when *ab initio* Patterson deconvolution techniques are applied.

from zero. This behaviour would make EDM procedures less effective.

A different trend is in general observed for structures with Wilsonian $\langle R^4\rangle$ moments. An example is shown in Fig. 3 for the protein with PDB code 1a0m [Hu *et al.* (1998), space group $I4$, resolution 1.09 Å], for which $\langle R^4\rangle = 2.02$; the increase of the σ_A parameter at decreasing values of the phase error is progressive and gradual. σ_A attains values close to unity only when the phase error is significantly small.

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References

Benach, J., Neely, H., Seetharaman, J., Ho, C. K., Janjua, H., Cunningham, K., Ma, L., Xiao, R., Liu, J., Baran, M. C., Acton, T. B., Rost, B., Montelione, G. T., Hunt, J. F. & Tong, L. (2013). *Northeast Structural Genomics Consortium*. In preparation.

Burla, M. C., Caliandro, R., Camalli, M., Carrozzini, B., Cascarano, G. L., Giacovazzo, C., Mallamo, M., Mazzone, A., Polidori, G. & Spagna, R. (2012). *J. Appl. Cryst.* **45**, 357–361.

Burla, M. C., Carrozzini, B., Cascarano, G. L., Comunale, G., Giacovazzo, C., Mazzone, A. & Polidori, G. (2012). *Acta Cryst.* A**68**, 513–520.

Burla, M. C., Carrozzini, B., Cascarano, G. L., Giacovazzo, C. & Polidori, G. (2011). *J. Appl. Cryst.* **44**, 1143–1151.

Burla, M. C., Carrozzini, B., Cascarano, G. L., Giacovazzo, C. & Polidori, G. (2012). *J. Appl. Cryst.* **45**, 1287–1294.

Burla, M. C., Giacovazzo, C., Mazzone, A., Polidori, G. & Siliqi, D. (2011). *Acta Cryst.* A**67**, 276–283.

Caliandro, R., Carrozzini, B., Cascarano, G. L., De Caro, L., Giacovazzo, C., Moustiakimov, M. & Siliqi, D. (2005). *Acta Cryst.* A**61**, 343–349.

Cascarano, G., Giacovazzo, C. & Luić, M. (1985). *Acta Cryst.* A**41**, 544–551.

Cascarano, G., Giacovazzo, C. & Luić, M. (1987). *Acta Cryst.* A**43**, 14–22.

Dauter, Z. & Adamiak, D. A. (2001). *Acta Cryst.* D**57**, 990–995.

Esposito, L., Vitagliano, L., Sica, F., Sorrentino, G., Zagari, A. & Mazzarella, L. (2000). *J. Mol. Biol.* **297**, 713–732.

Harata, K. (1994). *Acta Cryst.* D**50**, 250–257.

Hauptman, H. (1982). *Acta Cryst.* A**38**, 289–294.

Horvath, A. J., Irving, J. A., Law, R. H., Rossjohn, J., Bottomley, S. P., Quinsey, N. S., Pike, R. N., Coughlin, P. B. & Whisstock, J. C. (2013). In preparation.

Hu, S. H., Loughnan, M., Miller, R., Weeks, C. M., Blessing, R. H., Alewood, P. F., Lewis, R. J. & Martin, J. L. (1998). *Biochemistry*, **37**, 11425–11433.

Lunin, V. Yu. & Urzhumtsev, A. G. (1984). *Acta Cryst.* A**40**, 269–277.

Luzzati, V. (1952). *Acta Cryst.* **5**, 802–810.

Murshudov, G. N., Vagin, A. A. & Dodson, E. J. (1997). *Acta Cryst.* D**53**, 240–255.

Pan, B., Xiong, Y., Shi, K., Deng, J. & Sundaralingam, M. (2003). *Structure*, **11**, 815–823.

Read, R. J. (1986). *Acta Cryst.* A**42**, 140–149.

Shao, C., Zhang, F., Kemp, M. M., Linhardt, R. J., Waisman, D. M., Head, J. F. & Seaton, B. A. (2006). *J. Biol. Chem.* **281**, 31689–31695.

Sim, G. A. (1959). *Acta Cryst.* **12**, 813–815.

Srinivasan, R. & Ramachandran, G. N. (1965). *Acta Cryst.* **19**, 1008–1014.

Vlassi, M., Dauter, Z., Wilson, K. S. & Kokkinidis, M. (1998). *Acta Cryst.* D**54**, 1245–1260.